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(71) Applicant
**Novo Industri A/S (Denmark),
Novo Alle, DK—2880 Bagsvaerd, Denmark**

(72) Inventor
Kurt Albert Dorreich

(74) Agent and/or Address for Service
**Forrester Ketley & Co., Forrester House, 52 Bounds
Green Road, London N11 2EY**

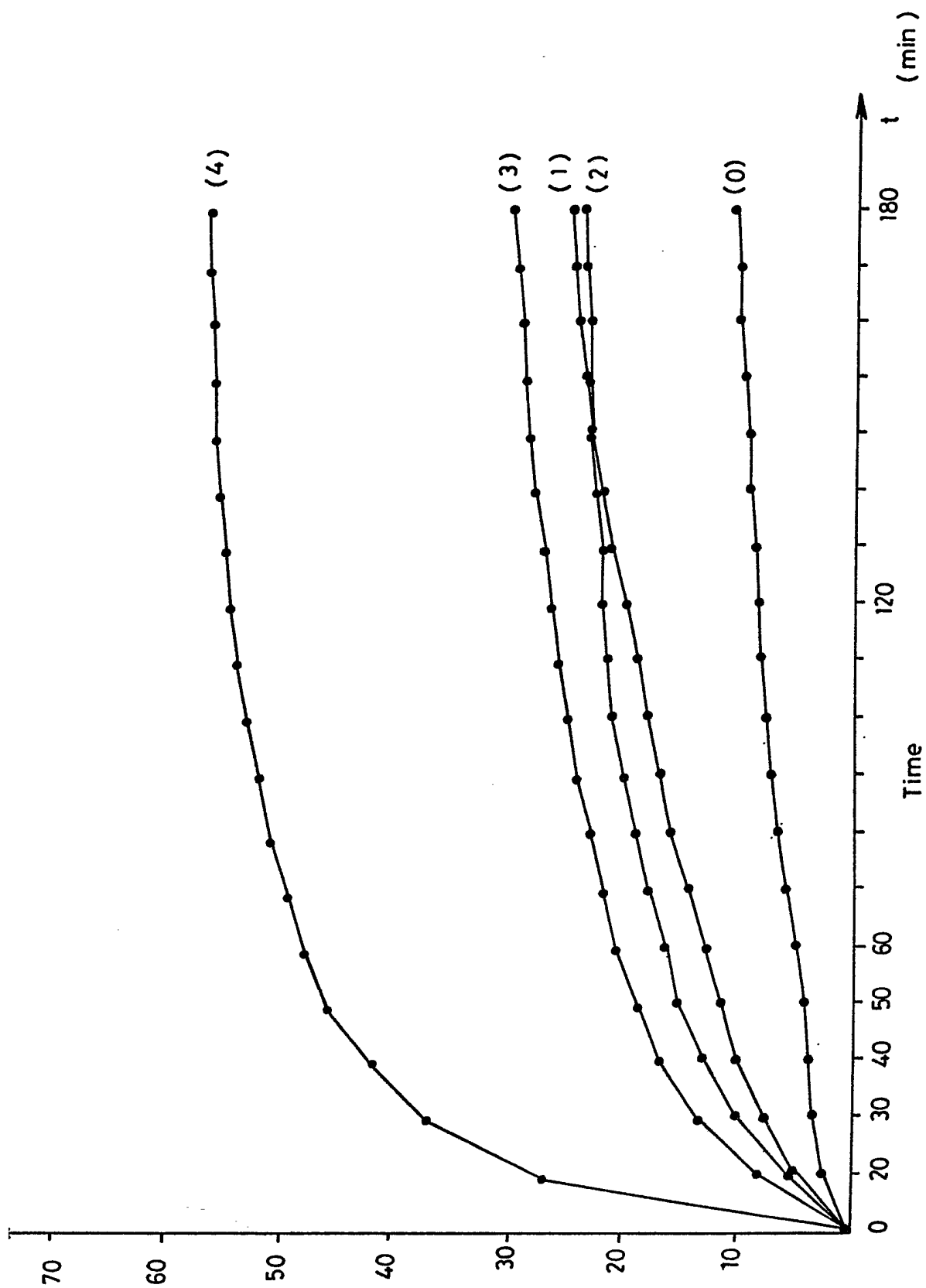
(54) **Method of enzymatic treatment of predejuiced mash**

(57) Predejuiced mash originating from fruit(s) or vegetable(s), is treated by addition of an aqueous medium to the predejuiced mash in order to form a viscous mass, and addition of an enzyme preparation comprising cellulase(s), hemicellulase(s) and pectinase(s) to the viscous mass, wherein the enzyme preparation is an SPS-ase (soluble polysaccharide-ase) preparation as described and/or claimed in UK Patent Application No. 8236309.

Preferably the SPS ase is obtained from *Asp. aculeatus* CBS 101-43.

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SPECIFICATION

Method of enzymatic treatment of predejuiced mash

The present invention relates to a method of enzymatic treatment of predejuiced mash.

Predejuiced mash is partially dejuiced mash, that is mash from which a part of the juice has been removed, preferably by pressing. 5

The invention comprises a method for enzymatic treatment of predejuiced mash originating from fruit(s) or vegetable(s), by addition of an aqueous medium in order to form a viscous mass, and by addition of an enzyme preparation comprising cellulase(s), hemi-cellulase(s) and pectinase(s) to the viscous mass.

10 Fruit and vegetable juices, especially apple juice, are produced mainly by pressing or by total liquefaction. Even if total liquefaction is the simplest way of obtaining a juice, the total liquefaction method requires a rather high amount of enzyme and, for that reason, the less enzyme demanding, conventional pressing method is often preferred.

By use of the conventional pressing method, a relatively high amount of predejuiced mash is generated. In US Patent Specification No. 4,275,648, it has been suggested to subject the predejuiced mash to an enzymatic treatment in order to generate some more juice from the predejuiced mash. However, with the conventional enzymes indicated in US Patent Specification No. 4,275,648, it has been found that the treatment of the predejuiced mash is relatively long, that the viscosity decrease rate of the viscous mass consisting of predejuiced mash and aqueous medium is relatively low, and that the throughput in relation to the pressing operation subsequent to the treatment of the predejuiced mash is relatively low. 15 20

Thus, the object of the present invention is to provide a method of enzymatic treatment of predejuiced mash, of the above indicated kind, by means of which the treatment time of the predejuiced mash can be significantly reduced the viscosity decrease rate of the viscous mass consisting of predejuiced mash and aqueous medium can be significantly increased and the throughput in relation to the pressing operation subsequent to the treatment of the predejuiced mash can be significantly increased. 25

Now, according to the invention, surprisingly it has been found that the above indicated object can be fulfilled, if a special enzyme preparation is used for the enzymatic treatment.

30 In accordance with the present invention, there is provided a method of enzymatic treatment of predejuiced mash originating from fruit(s) or vegetable(s) by addition of an aqueous medium in order to form a viscous mass and by addition of an enzyme preparation comprising cellulase(s), hemicellulase(s) and pectinase(s) to the viscous mass, wherein the enzyme preparation is an SPS-ase as described and/or claimed in UK Patent Application No. 8236309 (published under number 2115820A).

35 The aqueous medium could for example be tap water, deionized water or extraction liquid from a previous pressing of mash. 35

In a preferred embodiment of the method according to the invention, the SPS-ase is added in an amount corresponding to a MOU activity of from 10 to 2000 MOU units per kg of predejuiced mash (the MOU unit is defined hereinafter). In this manner, a compromise between low treatment time and high viscosity decrease rate can be obtained. 40

In another preferred embodiment of the method according to the invention, the added SPS-ase preparation is producible on the basis of *Aspergillus aculeatus* CBS 101.43. In this manner, an optimal ratio between cellulases, hemicellulases, pectinases and SPS-ase is obtained in regard to maximum decomposition.

45 In the method according to the invention, it is preferred that the treatment temperature is the range of from 10°C. to 65°C., advantageously from 15°C. to 30°C. or from 45°C. to 55°C. In the advantageous lower temperature range, the reaction rate is relatively low but, as compensation, the taste of the degradation product of the predejuiced mash is excellent. In the advantageous higher temperature range, the reaction rate is relatively high, whereas the taste of the degradation product of the predejuiced mash may be somewhat impaired; as the degradation product of the predejuiced mash is only a small fraction of the primary juice, this possible impairment is normally of no practical significance. The intermediate temperature range is less preferred due to risk of microbial contamination. 50

In the method according to the invention, it is advantageous that the treatment time is in the range of from 10 minutes to 15 hours, preferably from 30 minutes to 3 hours. In the lower end of the time interval, the decomposition of the predejuiced mash is relatively low but, as a compensation, the throughput through the plant is relatively high. In the higher end of the time interval, the decomposition of the predejuiced mash is relatively high, whereas the throughput through the plant is relatively low. 55

60 In a preferred embodiment of the method according to the invention, the pH during treatment is the natural pH and deionized water is used as the aqueous medium. In this manner, no interference with the composition of the natural mineral salts is generated. In another preferred embodiment of the method according to the invention, the extraction liquid from a previous pressing of mash is used as the aqueous medium. In this manner, the cost of final concentration of the liquid extract from the 60

predejuiced mash can be reduced.

The present invention is suited, for example, for use with predejuiced mash from apples, pears, black or red currants, peaches, apricots, berries, grapes, citrus fruits, tropical fruits, carrots, potatoes, celery, paprika, peas, tomatoes, beans, cabbage, or onions.

5 Usually, the dry weight of the pomace is in the general range of from 10 to 50% of the dry weight of the predejuiced mash used as a starting material. Thus, pomace is the fruit or vegetable residue, from which practically no more juice can normally be obtained. 5

By means of the present invention, an increase of the Brix yield in the final juice can be obtained by solubilization of polysaccharides other than pectins, due to the SPS-ase activity.

10 As appears from the following Examples, the present invention opens up the possibility of using a low enzyme dosage compared with conventional enzymes on an equal MOU-basis, 10

In the method described in US Patent Specification No. 4,275,648, the pomace resulting from the enzymatic treatment of the predejuiced mash is separated from the supernatant by means of centrifugal or gravitational forces. However, such devices are no part of normal plants for production of apple juice.

15 According to the present invention, no such separation devices are needed, because the same press, which was used in relation to the formation of the predejuiced mash used as the starting material in the method according to the present invention, can be also used for the separation of the pomace from the supernatant. Thus, the present invention provides the further technical advantage in comparison to the US Patent Specification No. 4,275,648 that the press can be used for the above mentioned dual 15

20 purpose, whereby investment costs in the juice production plant can be kept to a minimum. 20
The viscosity decrease rate with conventional enzymes and with an SPS-ase preparation according to the invention is compared in the following series of tests. In these tests, the Pectinex (prior art pectinase), other prior art enzymes and the SPS-ase preparation are used in corresponding concentrations, that is concentrations which generate the same pectinase activity in MOU units/kg of predejuiced mash, whereby the MOU activity is measured according to the "Determination of the 25

25 Pectinase Units on Apple Juice (MOU)" of 12.6.1981, available from Schweizerische Ferment AG, Vogesenstrasse 132, Basle, Switzerland. 25
Predejuiced apple mash which had been produced in the following way was used as a substrate: Apples were coarsely milled with a Bucher Central mill (4 mm). The apple mash was pressed until 75% of juice (weight/weight) was obtained. The resulting predejuiced apple mash was suspended in twice the amount of water and then milled on a Fryma mill with a coround stone outfit and a fissure of 0.5 mm. 30

30 Enzyme reactions were carried out at 50°C for 3 hours in the Contraves Epprecht Rheomat 15. During stirring, continuous viscosity measurements were carried out and the viscosity expressed as a percentage of the original viscosity was determined (speed setting 15). Table 1 and Figure 1 show a comparison between the effect of Pectinex 3X (2550 MOU/g, Celluclast 1.5 L, the combination of 35

40	SPSU/g	=	40	40
	SRU/g	=	205	
	PGE/g	=	9560	
	HUT at pH 3,2/g	=	3200	
	VHCU/g	=	256,000	

45 The results appear from the following Table 1 and Figure of the accompanying drawing. 45

TABLE 1: Viscosity decrease of suspension of predejuiced apple mash in percent
(Comparison of enzyme on equal MOU basis; Epprecht Rheomat 15 speed setting 15)

Time (min)	Blank (0)	Pectinex 3X (1)	Celluclast 1.5 L (2)	Pectinex 3X + Celluclast 1.5 L (3)	SPS-ase Preparation (4)
0	0	0	0	0	0
0	2,5	5,0	6,0	8,0	27,0
20	3,5	7,5	10,0	13,5	37,0
30	4,0	10,0	13,0	17,0	42,0
40	4,5	11,5	15,5	19,0	46,0
50	5,0	13,0	16,5	21,0	48,0
60	6,0	14,5	18,0	22,0	49,5
70	6,5	16,0	13,0	23,0	51,0
80	7,0	17,0	20,0	24,5	52,0
90	7,5	18,0	21,0	25,0	53,0
100	8,0	19,0	21,5	26,0	54,0
110	9,0	20,0	22,0	26,5	54,5
120	9,5	21,0	22,0	27,0	55,0
130	9,0	22,0	22,5	28,0	55,5
140	9,0	23,0	23,0	28,5	56,0
150	9,5	23,5	23,0	29,0	56,0
160	10,0	24,0	23,0	29,0	56,0
170	10,0	24,5	23,5	29,5	56,5
180	10,5	25,0	23,5	30,0	56,5

Appendix to Table 1

(0) : blank

(1) : 30 g of Pectinex 3X per 100 kg of predejuiced mash

(2) : 60 g of Celluclast 1.5 L per 100 kg of predejuiced mash

(3) : 30 g of Pectinex 3X and 60 g of Celluclast 1.5 L per 100 kg of predejuiced mash

(4) : 77.4 g of SPS-ase preparation per 100 kg of predejuiced mash

Pectinex 3X : 2,550 MOU/g

SPS-ase preparation : 988 MOU/g

The following Examples further illustrate the present invention.

EXAMPLE 1

Treatment of predejuiced apple mash originating from stored apples (Jonathon and Boskop:

5 Apples were milled with a Bucher Central mill (4 mm) and the mash pressed after a mass treatment with Pectinex (20 g Pectinex 1X per 100 kg apples, one hour, room temperature) with a Bucher horizontal press until 78% of juice was obtained.

0.667 kg of the resulting apple mash was suspended in 1,333 kg of water, heated to 50°C. The above indicated SPS-ase preparation was added (10 g per 100 kg of predejuiced mash) during stirring. After combined stirring for 2 hours, the mash was pressed with a Hafico HP5M press. 1 510 kg of "juice" with 5.5° Brix was obtained.

5 EXAMPLE 2

5

Industrial treatment of predejuiced apple mash.

To 6000 kg of predejuiced apple mash, which was received from a conventional processing fruit juice plant (Bucher Central mill: 6 mm; mash treatment: 10 g Pectinex 1X per 100 kg of apples, Bucher HP 5000 press) 12,000 l of hot water (condensate from the concentrator) was added and a mixing
10 temperature of 55°C was achieved. Then, 1.2 kg of the above indicated SPS-ase preparation was added. After two hours of slight stirring, the mass was pressed with a Bucher HP 5000 press, similar to
10 a press used for processing normal apple mashes. The weight of the pomace was 3700 kg, and
14,300 l of juice with 4.5°Brix was obtained.

10

The following Examples 3 to 12 were carried out generally as Example 2, but with the parameters
15 which appear from the following Table.

15

Example	Equipment for production of PDM*) Predejuiced mash	% of PDM*) in relation to weight of raw material	Quantity of PDM*), kg	Quantity of water, kg	Temperature of viscous mass, °C	Dosage of SPS-ase preparation, g/t PDM*	Enzymatic treatment time minutes	Stirring time, minutes	Separation equipment	Quantity of pomace, kg	Quantity of juice, kg	Juice concentration, °Brix
3	horizontal press (Bucher HP 5000)	75	6120	14 000	50	200	240	0	horizontal press (Bucher HP 5000)	3280	16 840	4,4
4	horizontal press (Bucher HP 5000)	70	6000	4000	27	200	65	65	horizontal press (Bucher HP 5000)	3400	6600	5,4
5	horizontal press (Bucher HP 5000)	83,3	12 280	25 000	45	200	180	180	horizontal press (Bucher HP 5000)	3680	33 600	4,5
6	horizontal press (Bucher HP 5000)	75	1695	2800	36	300	120	120	decanter (Westfalia)	1280	3215	4,6
7	horizontal press (Bucher HP 5000)	75	1890	3500	40	200	120	10	decanter (Westfalia)	1468	3922	4,9
8	belt press (Ensink)	68	31,9	61	50	400	90	90	belt press (Ensink)	13,8	79,1	4,7
9	belt press (Willmes Continupak)	68,3	990	1800	58	400	90	90	belt press (Bellmer angle press)	390	2400	4,0
10	belt press (Willmes Continupak)	69	1500	2000	56	400	90	90	belt press (Willmes Continupak)	1150	2350	4,5
11	belt press (Bini)	68	4500	1500	40	100	90	0	horizontal press (Bucher HP 5000)	1480	7020	6,3
12	belt press (Bini)	50	5000	1000	20	100	90	90	horizontal press (Bucher HP 5000)	1200	4800	8,8

In the foregoing, the words "Cellucast", "Pectinex", "Hafico", "Bucher", "Fryma" and "Contraves Epprecht" are Trade Marks.

CLAIMS

1. A method of enzymatic treatment of predejuiced mash originating from fruit(s) or vegetable(s), which method comprises addition of an aqueous medium to the predejuiced mash in order to form a viscous mass, and addition of an enzyme preparation comprising cellulase(s), hemicellulase(s) and pectinase(s) to the viscous mass, wherein the enzyme preparation is a SPS-ase preparation as described and/or claimed in UK Patent Application No. 8236309. 5
2. A method according to Claim 1, wherein the SPS-ase preparation is added in an amount corresponding to a MOU activity of from 10 to 2000 MOU units per kg of predejuiced mash.
3. A method according to Claim 1 or 2, wherein the added SPS-ase preparation is producible on the basis of *Asp. aculeatus* CBS 101.43. 10
4. A method according to Claim 1, 2 or 3, wherein the treatment temperature is in the range of from 10°C. to 65°C.
5. A method according to Claim 4, wherein the treatment temperature is in the range of from 15 to 30°C.
6. A method according to Claim 4, wherein the treatment temperature is in the range of from 45 to 55°C. 15
7. A method according to any one of Claims 1 to 6, wherein the treatment time is in the range of from 10 minutes to 15 hours.
8. A method according to Claim 7, wherein the treatment time is in the range of from 30 minutes to 3 hours. 20
9. A method according to any of Claims 1 to 8, wherein the pH during treatment is the natural pH and deionized water is used as the aqueous medium.
10. A method according to any one of Claims 1 to 8, wherein extraction liquid from a previous pressing of mash is used as the aqueous medium.
11. A method according to any one of Claims 1 to 10, wherein the fruit(s) or vegetable(s) is/are apples, pears, black or red currants, peaches, apricots, berries, grapes, citrus fruits, tropical fruits, carrots, potatoes, celery, paprika, peas, tomatoës, beans, cabbage or onions. 25
12. Fruit and/or vegetable juice produced using a method in accordance with any one of the preceding claims.
13. Any novel feature or combination of features described herein. 30

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INVENTOR-INFORMATION:

NAME	COUNTRY
DORREICH, KURT ALBERT	N/A

ASSIGNEE-INFORMATION:

NAME	COUNTRY
NOVO INDUSTRI AS	N/A

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ABSTRACT:

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Predejuiced mash originating from fruit
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of an aqueous medium to the
predejuiced mash in order to form a
viscous mass, and addition of an

enzyme preparation comprising cellulase (s), hemicellulase(s) and pectinase(s) to the viscous mass, wherein the enzyme preparation is an SPS-ase (soluble polysaccharide-ase) preparation as described and/or claimed in UK Patent Application No. 8238309. Preferably the SPS ase is obtained from *Asp. aculeatus* CBS 101.43. □ferably the SPS ase